Nontarget Impact of Spinosad GF-120 Bait Sprays for Control of the Mexican Fruit Fly (Diptera: Tephritidae) in Texas Citrus

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ABSTRACT Bait sprays containing the toxicant spinosad (GF-120) were applied to citrus groves in the Rio Grande Valley of Texas where Mexican fruit flies were detected in surveillance traps. The sprays were applied as a supplement to a continuous sterile insect release program. Sterile fly captures were 47–63% lower in the treated groves compared with control groves. Eight of 10 secondary pest populations declined in the test groves subsequent to spray applications, but they also declined in the control groves, suggesting that the decline was a seasonal phenomenon rather than a result of the bait sprays. Citrus whitefly, *Dialeurodes citri* (Ashmead), populations increased modestly and citrus blackfly, *Aleurocanthus woglumi* (Ashby), populations remained unchanged compared with pretreatment levels. Thus, no outbreaks of secondary pests occurred as a result of the spinosad bait sprays in this instance, as has been reported for malathion bait sprays in citrus. The bait sprays had no detectable effect on populations of specific indicator species of parasitoids (including *Aphytis* spp. and *Comperiella bifasciata* Howard), or on numbers of beneficial insects in general, in the treated groves.

KEY WORDS Secondary pests, bait sprays, *Anastrepha*

POPULATIONS OF THE MEXICAN fruit fly, *Anastrepha ludens* (Loew), are suppressed in southern Texas by an intensive sterile insect technique (SIT) program administered jointly by USDA–APHIS and the Texas Department of Agriculture. Flies are mass reared at a facility in Mission, TX, radiosterilized, and then dispersed by aircraft over the citrus growing areas of the lower Rio Grande valley at a rate of 30 million flies per week. In spite of these efforts, Mexican fruit flies are detected every spring in Texas in numbers sufficient to trigger export restrictions. Under existing quarantine protocols (Nilakhe et al. 1991), growers have the option of mitigative measures allowing export, which include application of pesticides to the infested groves. Until recently, the pesticide of choice for these applications was a Malathion bait spray (Lopez et al. 1969).

Generally, applications of insecticidal chemicals are incompatible with biological control programs. However, SIT, a form of biological control, is thought to be most, or only, efficacious when the sterile to fertile ratio is high (Knipling 1960). The ratio can be enhanced by either increasing the numbers of sterile flies released or by reducing the pest population with a supplementary control procedure. More than 350 Mexican fruit fly larvae can be reared from a single grapefruit (Thomas 1997); thus, the sterile fly capacity can be quickly overwhelmed by a local infestation, even when augmentative releases are applied to the focus of the outbreak. Chemical treatments are density independent and can knock down a localized infestation and thus enhance the sterile:fertile ratio.

A concern with pesticide applications is that they may induce an outbreak of secondary pests by the inadvertent suppression of natural enemies. In Texas, drift of methyl parathion from cotton fields has caused outbreaks in citrus of brown soft scale, *Coccus hesperidium* L., which is usually controlled by parasitic Hymenoptera (Dean et al. 1983). Such problems can be minimized by targeting the pesticide to the intended pest. By mixing the pesticide with a food-bait attractive to fruit flies and applying the formulation in droplets, a spray containing ultra-low volumes of pesticide can be applied (Prokopy et al. 1992). Vargas et al. (2002) reported that the Mediterranean fruit fly parasitoid *Fopius arisanus* (Sonan) would not consume the protein bait used in fly control programs. Nonetheless, such bait sprays targeting the Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), in California and Florida using malathion as the toxicant had negative impacts on pollinators and predatory insects within the eradication zone (Harris et al. 1980, Troetschler 1983, Hoelmer and Dahlsten 1993). Spinosad is a microbial metabolite with insecticidal properties that kills if ingested (Cisneros et al. 2002, Williams et al. 2003), but unlike malathion, it has little contact toxicity and therefore should have less impact on nontargets (Vargas et al. 2001). A spinosad formulation

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using mazoferm as the bait was found to be effective against Mediterranean fruit fly in Hawaii (Peck and McQuate 2000). Field trials in Florida demonstrated the efficacy of GF-120, a foliar toxic bait spray that combines spinosad and the lure solbait (Moreno and Mangan 2000) against the Mediterranean fruit fly and the Caribbean fruit fly, *Anastrepha suspensa* (Loew). But there were insufficient data to assess the effect on nontarget insects (Burns et al. 2001).

During the 2001 and 2002 citrus harvest seasons, the Texas fruit fly suppression program instituted experimental applications of GF-120, treating the first 10 locations where fertile Mexican fruit flies were detected in both years. We used this opportunity to measure the impact of GF-120 sprays on the populations of secondary pests, and their natural enemies, in the treated groves. We also report on the effect of these sprays on the sterile released Mexican fruit flies.

Materials and Methods

GF-120 is a mixture of the toxicant spinosad (DowAgroSciences, Indianapolis, IN) and a feeding attractant. The lure component, a hydrolyzed protein bait that attracts and induces feeding by fruit flies, was developed by Moreno and Mangan (1995). The basic formulation is derived from Mazoferm E802 (Corn Products, Argo, IL) an enzymatically hydrolyzed protein from corn processing. Liquid Mazoferm was shown to have no significant impact against a series of beneficial hymenopteran parasites by Dowell (1997) and reduced impact against honey bees (Dominguez et al. 2003). A dried and purified derivative from Mazoferm, Solulys, was developed and also successfully tested with a series of toxicants. The final bait formulation solbait uses solulys as the proteinaceous component. Additional components, and methods used in developing the formula, were described by Moreno and Mangan (2002). The solbait formulation contains 1% ammonium acetate (Sigma, St. Louis, MO), 1% polyethylene glycol 200 (ICN Biomedicals, Aurora, OH), 1% polysorbate 60 (Soco-Lynch, Los Angeles, CA), 0.25% soybean oil, 15% invertose (Liquid Sugar Inc., Emeryville, CA), 2% active ingredient (AI) Solulys, 0.4% xanthan gum, and ≈80% water. Spinosad has been shown to be highly toxic to fruit flies at lower concentrations than many commonly applied organophosphate insecticides (Chen et al. 2003, Stark et al. 2004). The label for GF-120 recommends 80 ppm concentration of spinosad in the solbait formulation (0.02%). For the tests described herein, the formulation was applied by aircraft (2001–2002) or by ground spray (2002–2003) at the recommended rate of 52 oz (1.5 liters) of mixture per acre.

The Texas Protocols specify that all citrus within 250 m of a fertile Mexican fruit fly detection be treated, which is an area of ≈12.5 ha. However, inasmuch as citrus groves in the lower Rio Grande valley are typically only 2 to 4 ha, a decision was made to treat groves in their entirety for this experiment, even though only a portion of the grove extended within the 250-m radius. The treated groves varied from as small as 0.25 ha to as large as 18 ha. In 2001–2002, the two smallest groves were treated manually using backpack sprayers. The rest were sprayed by airplane. In 2002–2003, all applications were made by ground using a ladybird sprayer mounted on an all terrain vehicle.

Table 1. Weather during treatment and sample dates for 2001–2002 season by week: rainfall (millimeters), maximum and minimum temperatures (°C), treatment (spray) dates (by grove number), nontarget insect sample dates (by grove number), and numbers of sterile flies released (in millions)

Wk	Rain	Max temp	Min temp	Spray	Sample	Sterile flies
Oct. 1–7	0	35	15			0
Oct. 8–14	0	34	16		1	21.2
Oct. 15–21	0	32	12	1, 2	2, 3	29.6
Oct. 22–28	0	36	15	3, 4	4	28.1
Oct. 29–Nov. 4	2	32	13	1, 2		24.8
Nov. 5–11	0	31	13	3, 4	5	28.3
Nov. 12–18	61	31	17	1, 2, 5		10.2
Nov. 19–25	1	30	10	3, 4		4.2
Nov. 26–Dec 2	1	32	3	1, 2, 5		36.1
Dec. 3–9	15	28	8	3, 4		26.7
Dec. 10–16	2	28	7	1, 2, 5		34.6
Dec. 17–23	8	28	7	3, 4		39.6
Dec. 24–31	6	26	2	5	6, 7	38.8
Jan. 1–7	1	23	−1	6, 7		43.7
Jan. 8–14	0	27	1	5		44.4
Jan. 15–21	14	29	7	6, 7		49.2
Jan. 22–28	0	32	6	8	8, 9	45.5
Jan. 29–Feb. 4	2	32	6	9		39.9
Feb. 5–11	4	29	6	8		26.9
Feb. 12–18	0	28	7	9		36.1
Feb. 19–25	5	32	7			40.3
Feb. 25–Mar. 4	5	29	4			32.4
Mar. 4–11	0	29	5			26.8
Mar. 12–18	0	34	9		2	30.7
Mar. 19–25	3	30	12		7	39.2
Mar. 26–April 1	0	32	18		1, 4	33.7
April 2–8	6	31	14		3	31.8
April 9–15	0	35	17		6, 8	33.5
April 16–22	0	33	23		9	34.7
April 23–30	0	36	22		5	26.4

In the 2001–2002 season, five successive sprays were applied to each treated grove at scheduled 14-d intervals. Because the harvest season for Texas citrus is October to March, these sprays were applied mainly in the winter. For example, the first fly detection was made on 9 October 2001. The first application was made on 17 October and the last on 14 December at this grove. For the last treated grove, applications began in January and the fifth application was made on 7 April. Table 1 provides the dates for each spray application in each test grove. Although program officials had planned to spray each of the first 10 groves where a fertile Mexican fruit fly was captured, one of the infested groves was a state-certified organic grove so the spray was not applied. At the time of the test, GF-120 had not yet been registered as an organic insecticide.

In the 2002–2003 season, four of the first 10 fly detections were in dooryards; thus, only six commercial groves received bait spray applications. The first grove received applications beginning in October 2002 and the sixth grove was treated beginning in February 2003. The treatment interval was reduced in this year to weekly sprays with the treatment dates for

Table 2. Weather during treatment and sample dates for 2002–2003 season by week: rainfall (millimeters), maximum and minimum temperatures (°C), treatment (spray) dates (by grove number), and sample dates (by grove number), and numbers of sterile flies released (in millions)

Wk	Rain	Max temp	Min temp	Spray	Sample	Sterile flies
Oct. 1–7	6	34	23			12.7
Oct. 8–14	1	34	17			6.3
Oct. 15–21	14	32	12			2.9
Oct. 22–28	111	33	15	1, 2		7.0
Oct. 29–Nov. 4	128	31	16	1, 2	1, 2	6.6
Nov. 4–11	0	31	9	1, 2	1, 2	8.2
Nov. 12–18	0	27	9	1, 2	1, 2	8.3
Nov. 19–25	28	27	11	1, 2	1, 2	21.4
Nov. 26–Dec. 2	0	23	3		1, 2	24.2
Dec. 3–9	0	27	3			30.6
Dec. 10–16	5	27	4			30.5
Dec. 17–23	0	29	2			32.4
Dec. 24–31	6	28	7			37.4
Jan. 1–7	0	28	3	3		30.2
Jan. 8–14	21	28	8	3	3	32.3
Jan. 15–21	0	27	2	3, 4, 5	3	30.5
Jan. 22–28	8	29	3	3, 4, 5	3, 4, 5	30.7
Jan. 29–Feb. 4	0	32	6	3, 4, 5	3, 4, 5	31.1
Feb. 5–11	12	26	3	4, 5	3, 4, 5	21.5
Feb. 12–18	0	32	5	4, 5, 6	4, 5	32.0
Feb. 19–25	5	27	2	6	4, 5, 6	30.3
Feb. 26–Mar. 4	13	26	2	6	6	31.5
Mar. 5–11	0	28	11	6	6	26.6
Mar. 12–18	0	31	17	6	6	30.7
Mar. 19–25	0	29	12		6	26.0
Mar. 26–April 1	0	29	5			29.6
April 2–8	0	32	15			27.8
April 9–15	0	29	9			28.4
April 16–22	0	31	21			30.5
April 23–30	0	35	21			30.0

each grove shown in Table 2. The interval was shortened because of concerns that rainfall could negatively impact the residual efficacy of the sprays. Weather data during the study periods of both years is provided in Tables 1 and 2. The National Weather Service station at Weslaco Texas is located on the grounds of the USDA laboratory and operated by USDA personnel. The cited data can be found in reports published by NOAA (2001–2003).

In the second year of study, the treated groves were scattered over three southern Texas counties so that none were in proximity to one another. In the first year of the study, three of the infested groves were located within the municipal boundaries of the same town, Weslaco, TX. In that instance, the closest pair was 3 km apart. By contrast, in all cases the control groves were within a 1-km radius of the corresponding treatment grove.

Secondary Pest Surveys. The damaging stages of the major pests of citrus are sessile or wingless. The reduced vagility, along with their aggregative behavior, greatly facilitated population sampling. We followed the survey methods for citrus pests described by Simanton (1962), modified for our situation. Before the spray applications, technicians collected a total of 100 leaves from each treatment grove, and 50 leaves from each of two control groves. The control groves were selected for their proximity to the treatment grove. These were always within 1 km of the test grove, and when there were options, control groves were chosen from opposite sides of the treatment grove. The leaf collection procedure consisted of sampling from 50 trees in the treatment grove and 25 trees in each of the control groves. Technicians entered each grove from opposite corners, beginning sampling at the second row from the end. Every other tree was sampled proceeding down rows until the quota was reached. Two leaves were collected blindly from each sampled tree, at shoulder height, one from outside and one from inside (arms-length) of the canopy. The two leaves from each tree were sealed in a petri dish for transport to the laboratory. Each leaf was scored in the laboratory for the presence of pest arthropods using a stereomicroscope. The leaves were scanned on the same day or held overnight in a refrigerator and scored the next day. Two months after the bait sprays, the treatment grove and the control groves were sampled again for comparison using the same procedures. The exact sampling dates are provided in Table 1.

Survey for Natural Enemies. Sticky traps are a standard method for sampling flying insects (Southwood 1978). Yellow sticky traps are the most attractive to parasitoids and predators in orchard studies (Dowell and Cherry 1981, Moreno et al. 1984, McClain et al. 1990). Before the bait spray application, sticky traps were placed in both treatment and control groves and retrieved after 4-d exposure. The traps employed were yellow, 10 by 20 cm, rectangular cards coated on both sides with adhesive (Gemplers, Bellville, WI), suspended from branches by a wire. The traps were hung inside the canopy at shoulder height in trees one row in from the corner at opposite sides of the grove. On removal from the grove the sticky trap was wrapped in cellophane to facilitate handling. All cards were subsequently examined under a stereomicroscope, and the predators and parasites identified and counted. This procedure was repeated 2 mo after the sprays to measure the effect, if any, on the populations of beneficial arthropods. The sticky cards were deployed during the same week that leaf samples were collected from the respective groves.

Trapping for Fruit Flies. Before, during, and after the bait spray applications, the program of sterile releases and detection trapping for fruit flies was in continuous operation. Plastic Multilure traps (Better World Manufacturing, Miami, FL) were baited with torula yeast suspended in 300 ml of water and 15 ml of propylene glycol, the latter as a preservative. Fruit flies removed from the traps were transported to the laboratory for identification and determination as feral or sterile. Before release, the SIT program flies were marked with a red fluorescent dye, visible under UV light. Also, just before release, the sterile flies were chilled to facilitate handling. The chilled flies (typically six million per load) were placed into a release machine mounted inside of a Cessna 206 aircraft. The release rate was 27,000 flies per km^2. Any unmarked flies were presumed to be wild flies and were dissected to verify reproductive status based on maturation of the testes or ovaries. The exact number of flies released varied somewhat from week to week averaging 30 million per week as shown in Tables 1 and 2.

In total, 2,200 traps were deployed for the suppression program with traps placed in host trees, about half of which were in commercial groves, and the other half in residential backyards. These traps were serviced weekly unless unfavorable weather conditions caused a delay in service. Each of the treatment groves and each of the control groves had a trap in continuous operation. After the initial 2001–02 test, we determined that this trap rate was insufficient to measure efficacy of the sprays; thus, in 2002–03 we increased the rate to four traps per grove. The traps were arrayed at compass point directions five rows away from the center or 10 trees away if in the same row. Furthermore, a synthetic lure containing two components, ammonium acetate and putrescine, marketed as Biolure (SUTERRA, Bend, OR), with water diluted [4:1 (vol:vol)] propylene glycol antifreeze (Prestone Lo-Tox) as the capture liquid, was deployed in the traps to increase trap efficiency.

Statistical Analysis. Data were analyzed using standard statistical tests (Sokal and Rohlf 1973). Student's t-test was used to compare mean numbers of secondary pests. The reported numbers of beneficial arthropods are the mean number captured on each of four sticky traps placed in each of the treatment groves or respective control groves. Model I analysis of variance (ANOVA) was used to compare mean numbers of beneficial arthropods between treated and nontreated groves and between posttreatment and pretreatment populations. Linear regression was used to correlate pest and predator populations. Probabilities for F and t statistics were computed using the online statistical program Unixstats (2005).

Results and Discussion

Impact of Spinosad Sprays on Natural Enemies. Because predators and parasites tend to be active, vagile organisms, substantial numbers are captured on sticky traps, thus providing a census method to compare populations in treatment and control groves. In total, 120 sticky traps were deployed in the groves before and after the spinosad treatments. The numbers of beneficial insects varied among the traps with a range from as few as four to as many as 1,217 individuals per trap. The total number of predators and parasitoids captured was 18,778. The mean number per trap was 156.5 ± 199.7. Although spiders, coccinellid beetles and lacewings are common in the citrus groves, the majority of the beneficials captured, 89.7%, were members of the Hymenoptera. Of these, 95.5% were Chalcidoidea.

Data in Table 3 suggest that there was no significant difference in populations of beneficial arthropods between the control (134.2 per trap) and treatment (132.1 per trap) groves before the sprays ($F = 0.0007$; df = 1, 16; $P = 0.98$). Populations of predators and parasites increased in all groves after the spray applications, a natural increase associated with the change in seasons (winter to spring). In that respect, a wintertime spray application may have the least impact on nontarget insects because of lower activity at that time. Comparing beneficial populations between treated and control groves after the sprays, the mean number of beneficial arthropods was 10% lower in the treatment groves (181.5 versus 210.7 per trap), possibly an effect of the spray applications, but the difference was not statistically significant ($F = 0.235$; df = 1, 16; $P = 0.63$). Moreover, the number of beneficial arthropods trapped was greater in four of the 10 treatment groves after the sprays than in the untreated controls.

Table 3. Mean number of beneficial insects (predators and parasites) per sticky trap by grove before and after spinosad bait spray applications

Grove no.	Pretreatment		Posttreatment	
	Treatment	Control	Treatment	Control
1	148.7	13.2	179.0	119.5
2	23.7	134.0	112.5	478.7
3	90.5	66.0	76.2	186.3
4	95.2	526.2	141.2	172.3
5	566.5	227.0	408.5	92.5
6	74.5	85.0	98.5	122.5
7	47.5	60.5	59.0	98.5
8	108.0	49.7	302.5	234.5
9	34.0	46.0	256.0	391.5
Mean	132.1a	134.2a	181.5a	210.7[a]
SD	167.5	153.5	117.8	129.1

Means followed by the same letter are not significantly different based on pairwise ANOVA. For treated groves, comparing before and after treatment: $F = 0.524$; df = 1, 16; $P = 0.48$.

To detect specific effects, we compared numbers of certain indicator species. *Comperiella bifasciata* Howard is an encyrtid introduced for control of the California red scale, *Aonidiella aurantii* (Mask). Before the sprays, *C. bifasciata* was about equally abundant in the control groves (7.2 ± 11.5) and treatment groves (5.9 ± 7.9) ($F = 0.08$; df = 1, 16; $P = 0.78$). After sprays, mean numbers trapped in the treated groves had dropped by ≈30% (4.1 ± 3.4), but this was not statistically significant ($F = 0.38$; df = 1, 16; $P = 0.55$). However, mean numbers increased in the nontreated groves by almost 100%, although again, the difference was not statistically significant ($F = 0.50$; df = 1, 16; $P = 0.49$). Factors that confounded the analysis were the nonuniform distribution of the wasp among the groves, which was correlated significantly with the incidence of its host ($r = 0.81$, $t = 3.87$, $P = 0.002$) and small total sample size ($n = 268$).

Among the commonest parasitoid wasps in the traps were species in the genus *Aphytis* (Aphelinidae) ($n = 4723$), which are natural enemies of chaff scale, *Parlatoria pergandii* Comstock (Woolley and Browning 1987), the commonest armored scale pest in the groves. Michaud (2003) demonstrated that GF-120 was attractive and caused significant mortality to *A. melinus* De Bach in cage tests, although it is much less lethal than the malathion-nulure formulation. In this case, the mean trap numbers of *Aphytis* increased in the treated groves, reaching 60.5 ± 64.2 after the sprays, compared with 46.9 ± 43.8 before the sprays.

Table 4. Incidence of secondary pests in treatment and control groves before and after spinosad sprays (mean ± SE% of leaves infested)

Pest	Treated grove		Control grove	
	Before	After	Before	After
Chaff scale	17.0 ± 10.4a	7.6 ± 5.11b	2.9 ± 7.2c	4.2 ± 10.8b
California red scale	10.2 ± 12.0a	3.1 ± 3.3b	6.1 ± 3.4c	2.5 ± 2.7b
Purple scale	4.1 ± 7.4a	2.4 ± 2.22a	0.6 ± 4.5a	2.7 ± 6.6a
Citrus black fly	9.2 ± 6.8a	9.0 ± 10.3ab	8.8 ± 4.7a	4.0 ± 3.3b
Citrus white fly	1.4 ± 1.9a	3.5 ± 8.7a	3.0 ± 3.7a	2.6 ± 4.1a
Wooley white fly	4.1 ± 3.0a	1.2 ± 1.0b	3.2 ± 3.4a	0.8 ± 0.8a
Citrus leaf miner	3.0 ± 3.0a	0.3 ± 0.5b	1.4 ± 1.3c	0.4 ± 1.0b
Texas citrus mite	16.1 ± 15.0a	14.1 ± 17.5a	16.0 ± 12.2a	11.5 ± 11.4a
Spider Mites	7.0 ± 4.8a	2.9 ± 3.7b	6.7 ± 5.4a	1.4 ± 1.6b
Citrus rust mite	4.3 ± 7a	03.8 ± 12.0a	5.4 ± 14.0a	1.6 ± 5.1a

Data are from nine treatment and 18 control groves. Means followed by the same letter are not significantly different at $P = 0.05$.

But this difference was not significant ($F = 0.278$; df = 1, 16; $P = 0.605$). In the control groves, the mean numbers actually dropped to 147.7 ± 321.6 *Aphytis* per trap in the spring compared with 269.7 ± 512.5 in the winter samples. Therefore, we did not detect a negative effect because of the spinosad treatments.

Impact of Spinosad Sprays on Secondary Pests. The most frequently encountered pests on Texas citrus are chaff scale; California red scale; citrus blackfly, *Aleurocanthus woglumi* (Ashby) (Aleyrodidae); and Texas citrus mite, *Anychus clarki* (McGregor). The citrus blackfly has been successfully controlled by an augmentative biological control program releasing a combination of two hymenopterous parasitoids, *Encarsia opulenta* Silvestri (Platygasteridae) and *Amitus hesperidum* Silvestri (Aphelinidae) (Summy and French 1988). In California, outbreaks of citrus blackfly have resulted from applications of malathion sprays for fruit flies (Ehler and Endicott 1984). Although no significant reduction in total numbers of predators or parasitoids was detected in our study, the secondary pest population was surveyed for impact that might be attributable to the sprays. In total, 10 pest species were sufficiently common to quantify (Table 4).

In the control groves, nine of the 10 pest species declined in incidence between the winter sample and the spring survey, concomitant with the increase in predator and parasite populations. Only the purple scale, *Lepidosaphes beckii* (Newman), population did not change (2.6 versus 2.7% of leaves infested). Similarly, in the treatment plots, the incidence of eight of the 10 pest populations declined. The citrus whitefly, *Dialeurodes citri* (Ashmead), more than doubled from 1.4 to 3.8%, but such low populations are not economically important. The citrus blackfly population did not change in the treated groves after the spray applications (9.9 versus 9.1% of leaves infested). Perhaps it could be argued that the lack of decline might be attributable to lesser control by beneficial arthropods considering that the populations in the nonsprayed groves declined by >50% (from 8.8 to 4.0%). Regardless, no outbreak of any pest species was induced by the spinosad sprays in this test.

Impact of Spinosad Bait Sprays on Sterile Fly Populations. In 2002, the first year of the spray program, a single fruit fly trap was in operation in each treated grove and in each adjacent nontreated grove. The captures of sterile flies from 5 wk of trapping in these groves during, and for 2 wk after, the last spray application was considered in the analysis. In the treatment groves, a mean of 26.9 ± 11.2 flies was captured per trap per week. During the same time period in the adjacent nontreated groves, the mean was 36.5 ± 25.5 flies per trap per week. Although populations were larger in the nontreated groves as expected, the difference was not statistically significant ($t = 1.28$, df = 22, $P = 0.107$). However, the design of the experiment contained a flaw that could have seriously affected the results. Program traps are placed on the periphery of the grove to facilitate access and to take advantage of the so-called "edge effect." Because sterile flies are released weekly, many of the flies captured in these traps were entering the groves and therefore were minimally subject to the treatments, if at all. Therefore, in the second year of the program the number of traps was increased, and these traps were arrayed in the interior of the groves.

In the second year of the program, four of the first 10 fertile Mexican fruit fly detections were in residential backyards, and sprays were restricted in those areas to potential host trees within a 200-m radius of the original find. Our experiment was thus limited to the six commercial groves that were treated. In all treated groves, there were fewer sterile flies captured than in the control groves and in some cases the difference was large. The mean reduction in numbers of Mexican fruit flies was 47.5%. Again, however, the difference was not statistically significant ($t = 1.28$, df = 5, $P = 0.13$) because of the great variation in numbers among groves (Table 5). Because fertile fly detections, and therefore the spray applications, are sequential, the replicates are not simultaneous, and therefore subject to different climatic conditions. Most notably, there was no detectable reduction in the sterile fly population in the first treated grove. Rains during late October–early November when the sprays were applied are suspected as a cause of reduced efficacy (Table 1). Removing that one treatment from

Table 5. Captures of sterile flies during GF-120 applications in treated and adjacent nontreated control groves (four traps each), 2003 season

Grove	Treated	Control
1 La Feria	140	145
2 Donna	6	7
3 Hargill	16	100
4 Citrus City	25	85
5 Edinburg	12	39
6 Perezville	73	129
Total	272	505
Mean	45.3 ± 52.3	84.2 ± 52.8
Test of means	$t = 1.28$, df = 5, $P = 0.13$	

the analysis results in a 63% difference in captures in the treated groves compared with the adjacent nontreated groves, and this difference was statistically significant ($t = 3.28$, df = 4, $P = 0.015$).

These results suggest that the sterile fly populations in the treated groves are reduced on the order of 50–60% during and immediately after the spray applications. Given the weekly replenishment of sterile flies and the continuous movement of sterile flies into the grove from nontargeted areas, the loss of steriles may be acceptable, provided the sprays are efficacious in reducing the fertile fly population in the same proportion.

Impact of Spinosad Bait Sprays on Wild Fertile Mexican Fruit Fly Populations. Subsequent to the spinosad sprays in the nine groves where feral flies were detected in 2002, recidivism occurred in three of the treated groves with six feral flies captured. By contrast, in the 18 adjacent, nontreated plots, nine feral flies were captured in four of the groves. Because a slightly higher frequency of feral flies was found in the treated compared with the nontreated groves, one might conclude that the spray had little benefit. But the comparison is biased because the treated groves were known to be infested, whereas the untreated control groves were presumably not infested, at least, at the time of the treatments. A better comparison might be between treated and untreated infested groves. In 2003 recidivism occurred in only one of the six treated groves ($n = 2$ flies). By comparison, the next six groves with fly detections were not treated and among these, feral flies were later captured in three of the six ($n = 5$ flies), suggesting that the treatments were effective. It is difficult to attach much significance to these results because the numbers of feral flies are so low, although the measured reduction, 40–60%, is on the same order of magnitude as the reduction in sterile fly populations.

Another way of considering the impact of the sprays is by examination of the total fertile fly population during the years in which the spray program was implemented. In 2002, the first year of the spray program, 715 wild Mexican fruit flies were captured valley-wide. In the second year of the program, the numbers fell to 305 flies captured. In the third year of the program, only 85 wild flies were captured. Inasmuch as the acreage of citrus, the numbers of traps deployed, and the numbers of sterile flies released, have all remained unchanged, the steady reduction in the numbers of wild flies during the implementation of the spray program is encouraging.

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References Cited

Burns, R. E., D. L. Harris, D. S. Moreno, and J. E. Eger. 2001. Efficacy of spinosad bait sprays to control Mediterranean and Caribbean fruit flies (Diptera: Tephritidae) in commercial citrus in Florida. Fla. Entomol. 84: 672–678.

Chen, H., B. McPheron, G. Krawczyk, and L. Hull. 2003. Laboratory evaluation of surface residue toxicity of insecticides to apple maggot flies. Int. Pest Control 45: 1–3.

Cisneros, J., D. Goulson, L. C. Derwent, D. I. Penagos, O. Hernandez, and T. Williams. 2002. Toxic effects of spinosad on predatory insects. Biol. Control 23: 156–163.

Dominguez, V. M., J. L. Leyva, D. S. Moreno, F. J. Trujillo, R. Alatorre, and A. Enrique Becerril. 2003. Toxicidad sobre *Apis mellifera* de cebos empleados en el combate de moscas de la fruta. Manejo Integrado Plagas Agroecologia 69: 66–72.

Dean, H. A., J. V. French, and D. Meyerdirk. 1983. Development of integrated pest management in Texas citrus. Publ. B-1434, Texas Agric. Exp. Sta., College Station, TX.

Dowell, R. V. 1997. Laboratory toxicity of a photo activated dye mixture to six species of beneficial insects. J. Appl. Entomol. 121: 271–274.

Dowell, R. V., and R. H. Cherry. 1981. Survey traps for parasitoids and coccinellid predators of the citrus blackfly, *Aleurocanthus woglumi*. Entomologia Experimentalis et Applicata. 29: 356–362.

Ehler, L. E., and P. C. Endicott. 1984. Effect of malathion-bait sprays on biological control of Insect pests of olive, citrus, and walnut. Hilgardia 52: 1–47.

Harris, E. J., A. Hafravoui, and B. Toulette. 1980. Mortality of nontarget insects by poison bait applied to control the Mediterranean fruit fly. Proc. Hawaiian Entomol. Soc. 23: 227–229.

Hoelmer, K. A., and D. L. Dahlsten. 1993. Effects of malathion bait spray on *Aleyrodes spiraeoides* (Homoptera: Aleyrodidae) and its parasitoids in northern California. Environ. Entomol. 22: 49–56.

Knipling, E. F. 1960. Use of insects for their own destruction. J. Econ. Entomol. 53: 415–420.

Lopez, F., D. L. Chambers, M. Sanchez, and M. Kamasaki. 1969. Control of the Mexican fruit fly by bait sprays concentrated at discrete locations. J. Econ. Entomol. 62: 1255–1257.

McClain, D. C., G. C. Rock, and J. B. Woolley. 1990. Influence of trap color and San Jose Scale (Homoptera: Diaspididae) pheromone on sticky trap catches of 10 aphelinid parasitoids (Hymenoptera). Environ. Entomol. 19: 926–931.

Michaud, J. P. 2003. Toxicity of fruit fly baits to beneficial insects in citrus. J. Insect Sci. 3: 1–9.

Moreno, D. S., and R. L. Mangan. 1995. Responses of the Mexican fruit fly (Diptera: Tephritidae) to two hydrolyzed proteins and incorporation of phloxine B to kill adults, pp. 257–259. *In* J. R. Heitz and K. Downum [eds.], Light activated pest control. ACS. Symposium Series 616. American Chemical Society, Washington, DC.

Moreno, D. S., and R. L. Mangan. 2002. A bait matrix for reduced-risk insecticides used against fruit flies (Diptera: Tephritidae), pp. 333–362. *In* G. Hallman and C. P. Schwalbe [eds.], Invasive arthropods in agriculture. Science Publishers Inc., Enfield, NH.

Moreno, D. S., W. A. Gregory, and L. K. Tanigoshi. 1984. Flight response of *Aphytis melinus* (Hymenoptera: Aphelinidae) and *Scirtothrips citri* (Thyasanoptera: Thripidae) to trap color, size and shape. Environ. Entomol. 13: 935–940.

Moreno, D. S., and R. L. Mangan. 2000. Novel insecticide strategies such as phototoxic dyes in adult fruit fly control and suppression programmes, pp. 421–432. *In* K. H. Tan [ed.], Area-Wide Control of Fruit Flies and other Insect Pests. Penerbit Universiti, Penang, Malaysia.

Nilakhe, S. S., J. N. Worley, R. Garcia, and J. L. Davidson. 1991. Mexican fruit fly protocol helps export Texas citrus. Subtrop. Plant Sci. 44: 49–52.

[NOAA] U.S. National Weather Service. 2001–2003. Climatological data: Texas. Vols. 106–108. National Climatic Data Center, Asheville, NC.

Peck, S. L., and G. T. McQuate. 2000. Field tests of environmentally friendly malathion replacements to suppress wild Mediterranean fruit fly (Diptera: Tephritidae) populations. J. Econ. Entomol. 93: 280–289.

Prokopy, R. J., D. R. Papaj, J. Hendrichs, and T. T. Wong. 1992. Behavioral responses of *Ceratitis capitata* flies to bait spray droplets and natural food. Entomol. Exp. Appl. 64: 247–257.

Simanton, W. A. 1962. Operation of an ecological survey for Florida citrus pests. J. Econ. Entomol. 55: 105–112.

Sokal, R. R., and F. J. Rohlf. 1973. Introduction to Biostatistics. W. H. Freeman, San Francisco, CA.

Southwood, T.R.E. 1978. Ecological Methods with particular reference to the study of insect populations, 2nd ed. Chapman & Hall, London, United Kingdom.

Stark, J. D., R. Vargas, and N. Miller. 2004. Toxicity of spinosad in protein bait to three economically important tephritid fruit fly species (Diptera: Tephritidae) and their parasitoids (Hymenoptera: Braconidae). J. Econ. Entomol. 97: 911–915.

Summy, K. R., and J. V. French. 1988. Biological control of agricultural pests: concepts every producer should understand. J. Rio Grande Hort. Soc. 41: 119–133.

Thomas, D. B. 1997. Degree-day accumulations and seasonal duration of the pre-imaginal stages of the Mexican fruit fly (Diptera: Tephritidae). Fla. Entomol. 80: 71–79.

Troetschler, R. G. 1983. Effects on nontarget arthropods of malathion bait sprays used in California to eradicate the Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae). Environ. Entomol. 12: 1816–1822.

Unixstats. 2005. http://ergo.ucsd.edu/cgi-bin/unixstats/probcalc. University of California, San Diego.

Vargas, R. I., S. L. Peck, G. T. McQuate, C. G. Jackson, J. D. Stark, and J. W. Armstrong. 2001. Potential for areawide integrated pest management of Mediterranean fruit fly (Diptera: Tephritidae) with a braconid parasitoid and a novel bait spray. J. Econ. Entomol. 94: 817–825.

Vargas, R. I., N. W. Miller, and R. J. Prokopy. 2002. Attraction and feeding responses of Mediterranean fruit fly and a natural enemy to protein baits laced with two novel toxins, phloxine B and spinosad. Entomol. Exp. Appl. 102: 273–282.

Williams, T., J. Valle, and E. Viñuela. 2003. Is the naturally derived insecticide Spinosad compatible with insect natural enemies? Biocontrol Sci. Technol. 13: 459–475.

Woolley, J. B., and H. W. Browning. 1987. Morphometric analysis of uniparental *Aphytis* reared from chaff scale, *Parlatoria pergandii* Comstock, on Texas citrus (Hymenoptera: Aphelinidae; Homoptera: Diaspididae). Proc. Entomol. Soc. Wash. 89: 77–94.

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